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Original Research Article

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Therapeutic Doses of Efzofitimod Demonstrate Efficacy in Pulmonary Sarcoidosis

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Take Home Message: This post hoc analysis of a phase 1b/2a trial, informed by results from an *in vitro* assay, shows that therapeutic doses of efzofitimod decreased relapses after tapering corticosteroids in patients with pulmonary sarcoidosis.

Keywords: sarcoidosis, granuloma, efzofitimod, corticosteroid, relapse

Ethics Statements: The studies were conducted in accordance with the Declaration of Helsinki and were reviewed and approved by the Central—Western Institutional Review Board-Copernicus Group, North Carolina (IRB00000533); Local—Cleveland Clinic, Ohio (IRB00000684); Medical University of South Carolina, South Carolina (IRB00001888); University of Illinois, Illinois (IRB00000083); University of Texas Southwestern Medical Center, Texas. The patients/participants provided their written informed consent to participate in this study.

Abstract

Background: In a phase 1b/2a clinical trial of efzofitimod in patients with corticosteroid-requiring pulmonary sarcoidosis, treatment resulted in dose-dependent improvement in key endpoints. We undertook a post hoc analysis pooling dose arms that achieved therapeutic concentrations of efzofitimod (Therapeutic group) versus those that did not (Subtherapeutic group).

Methods: Peripheral blood mononuclear cells incubated with tuberculin-coated beads were exposed to varying concentrations of efzofitimod in an *in vitro* assay to determine concentrations that inhibited granuloma formation. In the post hoc analysis, we compared time-to-first-relapse and changes in pulmonary function after a protocolized corticosteroid taper in the Therapeutic and Subtherapeutic groups.

Results: Efzofitimod at ≥300 nM (19 µg/ml) inhibited granuloma formation *in vitro*. Based on average efzofitimod serum concentrations achieved in the phase 1b/2a study, the 3 and 5 mg/kg dose arms were pooled as the Therapeutic group, while the 1 mg/kg arm was pooled with the placebo arm as the Subtherapeutic group. Relapse rates were 54.4% and 7.7% in the Subtherapeutic group and Therapeutic group, respectively. Median time-to-first-relapse in the Subtherapeutic group was 126 days, whereas in the Therapeutic group, only one of 17 patients relapsed by the end of the 24-week study (p=0.017). Slopes analysis showed that forced vital capacity increased in the Therapeutic group, but decreased in the Subtherapeutic group, over the course of the trial (p=0.035).

Conclusion: Treatment with efzofitimod at therapeutic doses, as compared with a subtherapeutic dose or placebo, was associated with a lower rate of relapse as corticosteroids were tapered.

Introduction

Efzofitimod (formerly ATYR1923; aTyr Pharma, Inc., San Diego, CA), a novel immunomodulatory agent currently in development for the treatment of interstitial lung disease, was found in a recent multiple ascending dose (1, 3, and 5 mg/kg) phase 1b/2a study to be safe and well-tolerated in patients with chronic pulmonary sarcoidosis on corticosteroids[1]. The study also showed a dose- and exposure (concentration)-dependent reduction of the mean daily dose of oral corticosteroid needed to maintain disease stability, with concomitant increases in quality of life scores and a trend toward improvement in pulmonary function[1,2]. The exposure-dependent response in the phase 1b/2a study implied that efzofitimod was most effective at higher doses, while less effective or not effective at lower doses. In drug development, the relationship between exposure and response is often established from *in vitro* assays. The current study was designed to examine the relationship between concentrations of efzofitimod that are effective *in vitro* and efficacy outcomes in sarcoidosis patients treated with different doses of the drug in the phase 1b/2a trial.

The hallmark of sarcoidosis is formation of non-caseating granulomas in the lungs and other affected tissues. Peripheral blood mononuclear cells (PBMCs) from patients with sarcoidosis can also form granulomas in culture, and this process can be quantified using an *in vitro* granuloma formation assay [3,4]. In the *in vitro* assay, PBMCs cultured in the presence of tuberculin purified protein derivative (PPD) aggregate and form multicellular structures with histopathologic and molecular features that closely resemble those of granulomas in human sarcoidosis tissues[5,6]. The assay provides a platform for testing the ability of drugs to inhibit granuloma formation *in vitro* and for determining concentrations at which a test drug may be efficacious *in vivo*. As such, the *in vitro* assay was recently used to predict the dose range expected to be effective in a clinical study of a monoclonal antibody being developed for treatment of sarcoidosis[4]. In the current study, we applied a similar approach to determine concentrations of efzofitimod that inhibit granuloma formation *in vitro*, and related the results to serum concentrations of the drug achieved with the doses used in the phase 1b/2a trial.

Here we present a post hoc analysis of the phase 1b/2a study demonstrating the favourable effects of treatment with efzofitimod at doses determined to be therapeutic based on the *in vitro* assay, as compared to a sub-therapeutic dose pooled with placebo, on relapse rates

and pulmonary function in subjects with chronic pulmonary sarcoidosis while they underwent a protocolized oral corticosteroid taper.

Methods

In vitro study

Blood for the *in vitro* assay was drawn under a protocol approved by The Ohio State University Institutional Review Board. The assay protocol was similar to that previously described[3,4]. All enrolled subjects (n = 8) had active pulmonary sarcoidosis, were non-smokers, had a negative tuberculin skin test and/or QuantiFERON-TB Gold test, and had not been treated with corticosteroids or other immunosuppressive medications (e.g. methotrexate, azathioprine, anti-TNF monoclonal antibodies) in the preceding 6 months. Sarcoidosis was deemed to be active based on the presence of intolerable symptoms, progressive lung dysfunction and/or high risk for other organ damage due to the disease, as assessed by one of the authors, who is a sarcoidosis specialist physician (E.D.C). Prior to initiation of treatment, blood was drawn for isolation of PBMCs. PBMCs were plated and cultured for 7 days in the presence of either uncoated polystyrene beads (UNC) or beads coated with tuberculin PPD[3,7]. In addition, PBMCs were treated with either vehicle or efzofitimod at 30 nM, 300 nM or 1 µM, or prednisone at 1 or 10 µM as positive control, for 30 min prior to addition of PPD-coated beads and throughout the subsequent 7-day culture period. After 7 days, granuloma formation was evaluated by light microscopy, analyzed using Materials Image Processing and Automated Reconstruction (MIPAR™ v2.2.5; Worthington, OH), and expressed as area fraction percent of uncoated beads, as previously described[3,4,7].

Statistical analysis – *in vitro* study

Data derived from independent experiments were expressed as boxplots. Statistical impact relative to sample size was further evaluated by employing Cohen's d effect size [8], which considers the magnitude of the change in the experimental value and the standard deviation of the measurements (i.e., a sensitivity index). A strong effect size is reflected by a Cohen's d value exceeding 0.8[8]. SigmaPlot 15.0 and SYSTAT 13.2 (Grafiti LLC, Palo Alto, CA) software were used for graphics and statistical analysis, respectively. The significance of differences in granuloma area

fraction among treatment groups was assessed with the Mann-Whitney U-test. Statistical significance was set at $p<0.05$.

Post hoc analysis of phase 1b/2a clinical trial

A post hoc analysis was performed on data from the previously reported phase 1b/2a study[1]. Briefly, the clinical study was a double-blind, placebo-controlled trial with three sequential ascending dose cohorts. Subjects were randomized 2:1 to receive either efzofitimod (1, 3, or 5 mg/kg in the first, second, and third cohorts, respectively) or placebo. The study enrolled symptomatic (Modified Medical Research Council [mMRC] Dyspnea Scale score ≥1) subjects with a ≥6-month history of biopsy-confirmed sarcoidosis[9], pulmonary parenchymal involvement detected by chest imaging, and treatment with oral corticosteroids (OCS) at a prednisone-equivalent dose of 10 to 25 mg/day without change for at least 4 weeks. Concomitant treatments (other than biologics)[10] for sarcoidosis were allowed and required to be maintained at a stable dose for the duration of the study. Subjects in each cohort received six doses of efzofitimod or placebo, administered intravenously at four-week intervals. During the course of the trial, subjects were required to decrease their OCS dose as outlined in a pre-specified taper protocol. A successful taper was defined as reduction of the OCS dose to a prednisone-equivalent of 5 mg per day or less for at least 5 consecutive days. The protocol allowed for return to a higher OCS dose as "rescue" therapy for increased symptoms (cough or dyspnea) judged by the investigator to represent significant clinical worsening. Time-to-first-relapse was defined as the interval from the date of the first successful OCS taper to the date when "rescue" therapy was first required. Increases in OCS dose for reasons other than worsening sarcoidosis were not counted as relapses.

The key efficacy parameters for the post hoc analysis were time-to-first-relapse and changes in pulmonary function. The pulmonary function parameters evaluated included forced vital capacity (FVC), forced expiratory volume at one second (FEV_1), and diffusing capacity of the lung for carbon monoxide (DLCO). Percent predicted values for FVC and FEV1 were determined using race/ethnicity-specific Global Lung Initiative (GLI) reference equations[11] based on patients' self-reported race/ethnicity. Percent predicted values for DLCO were determined using reference equations in place at each study center at the time of the trial.

Statistical analysis – post hoc study

Based on findings from the *in vitro* granuloma formation study (see Results below), the 3 and 5 mg/kg dose arms were pooled as the Therapeutic group, and the 1 mg/kg arm was pooled

with the placebo arm as the Subtherapeutic group. The primary efficacy analysis was in the modified intention-to-treat (mITT) population, defined as all randomized patients who received at least one administration of study drug or placebo. We compared the Therapeutic and Subtherapeutic groups for endpoints pre-specified in the Statistical Analysis Plan, including time-to-first-relapse after OCS taper, and changes in FVC, FEV_1, and DLCO percent predicted (DLCOpp) over the 24 weeks of the study. Time-to-first-relapse in the two groups was analysed with the Log-rank test and is presented as a Kaplan-Meier plot. Subjects who were not able to taper their OCS dose to 5 mg prednisone-equivalent or less during the study were included in the analysis as censored values on Day 1. Changes in FVC and FEV_1 over the course of the trial in the two groups were analysed using the random coefficient regression model (RCRM), as in the primary publication[1]. An interaction term was added to the RCRM to assess whether there was evidence that improvements in FVC were consistent for patients with differing values of FEV1/FVC ratio at baseline (data not shown). The change from baseline in DLCOpp at weeks 12, 20 and 24 was calculated for each individual and the significance of differences between treatment groups was analysed using a mixed-effects model for repeated measures. Baseline pulmonary function values were used as co-variates in the analysis. For all tests, statistical significance was set at $p<0.05$.

Results

In vitro study

Eight subjects were studied (Supplemental Table 1). The half maximal effective concentration (EC_{50}) for efzofitimod binding to its human receptor neuropilin 2[12] is 30 nM (1.9 µg/mL)[13]. Thus, we tested the effect of efzofitimod at 30 nM, 300 nM and 1 µM on granuloma formation in the *in vitro* assay. Treatment with efzofitimod at 30 nM (1.9 ug/mL) did not significantly affect granuloma formation; whereas treatment at 300 nM decreased granuloma area fraction by approximately 60% ($p<0.05$) (Figure 1). Based on the finding that efzofitimod at 300 nM (19 µg/mL) reduced granuloma formation *in vitro*, we considered this concentration as one that would potentially be therapeutically effective in patients with sarcoidosis.

Post hoc analysis of the Phase 1b/2a trial

The area under the efzofitimod concentration-time curve (AUC) over the 4-week (672-hour) dosing interval for the 1, 3, and 5 mg/kg dose groups was 3,710, 12,077, and 16,122

µg*hr/mL, respectively. The calculated average concentration (Cavg) over the dosing interval (AUC in µg*hr/ml divided by 672 hours) at 1 mg/kg (5.5 µg/mL) was less than the effective concentration in the *in vitro* assay, while that for the 3 mg/kg (18.0 µg/mL) and 5 mg/kg (24.0 µg/mL) doses was similar to or greater than the concentration that inhibited granuloma formation *in vitro* (19 µg/mL). Therefore, for the post hoc analysis, the 3 mg/kg and 5 mg/kg cohorts were considered to have received effective doses of efzofitimod and pooled as the Therapeutic group, while 1 mg/kg cohort arm was considered to have received a less than effective dose of the drug and pooled with the placebo cohort as the Subtherapeutic group.

a) Patient characteristics:

The phase 1b/2a study enrolled 37 subjects. Based on the justification for pooling described above, 20 were pooled in the Sub-therapeutic group, and 17 were pooled in the Therapeutic group. Demographics, disease characteristics, and baseline immunosuppressive therapies for subjects in the two groups are shown in Table 1. The mean age of study participants was 52.4 years, 54% were female, and 38% were African American. Pulmonary function parameters were notable for mild to moderate reductions in FVC, FEV_1, and DLCO in both the Subtherapeutic and Therapeutic groups. At enrolment, all patients were on oral corticosteroid therapy at prednisone-equivalent doses of 10 to 25 mg/day, and 14 of 37 (38%) were also on non-steroid immunomodulatory medications. The mean prednisone-equivalent dose at baseline was 13.2 mg, and at least 20% of patients in each group were on ≥20mg/day. Baseline measures were defined as the last measure assessed on or before the first efzofitimod (or placebo) dose. Overall, demographics, disease parameters, and baseline medications were similar in the two groups.

b) Efficacy assessments:

i) Relapse and time-to-first-relapse: Of the 37 randomized patients, 32 achieved a reduction of OCS to a prednisone-equivalent dose of ≤5 mg for at least 5 consecutive days. Of the 5 patients who were unable to taper to 5 mg or less, 3 were in the Subtherapeutic group and 2 were in the Therapeutic group. As shown in Figure 2, time-to-first-relapse was significantly shorter in the Subtherapeutic group than in the Therapeutic group. The median time-to-first-relapse in the Subtherapeutic group was 126 days, whereas only one of 17 patients in the Therapeutic group had relapsed by the end of the study (p=0.017). The relapse rate in the Subtherapeutic group was 54.4%, compared to 7.7% in the Therapeutic group (Table 2).

ii) FVC and FEV_1: A slopes analysis showed that over the course of the trial, FVC increased from

baseline in the Therapeutic group, but decreased from baseline in the Subtherapeutic group (p=0.035; Figure 3b). The mean FVC at the end of the study was 3165 mL in the Therapeutic group and 2985 mL in the Subtherapeutic group (mean difference 180 mL, p=0.035; Table 2). The results were similar when the data were analysed as FVC percent predicted (FVCpp; data not shown).

Slopes analysis also showed that FEV_1 increased in the Therapeutic group, but decreased in the Subtherapeutic group, over the course of the trial, although the trend did not reach statistical significance (p=0.196; Figure 4 and Table 2). This was also the case when the data were analysed as FEV_1 percent predicted (FEV_1pp; data not shown). There was no evidence that variation in baseline FEV_1/FVC was associated with differential effects of efzofitimod on FVC (p=0.466 [interaction test]).

iii) DLCO: We were unable to perform a slopes analysis for DLCO measurements over the course of the trial because DLCO was only measured at a few selected times during the trial. However, by the end of the 24-week study, the adjusted mean percent predicted DLCO (DLCOpp) was 7.4% greater in the Therapeutic group than in the Subtherapeutic group (Table 2). While this change favoured the Therapeutic group, the trend did not reach statistical significance (p=0.104).

Discussion

In the phase 1b/2a trial, efzofitimod was safe and well-tolerated, and treatment was associated with statistically significant dose-dependent improvement in patient-related outcomes assessed using multiple validated sarcoidosis-specific instruments[1]. Although the study was not powered to determine the drug's efficacy in maintaining disease control as OCS were tapered or in terms of pulmonary function, treatment was associated with dose-dependent trends toward improvement in these secondary endpoints. In addition, Walker et al recently performed an exposure-response analysis that revealed exposure-dependent trends supporting the efficacy of efzofitimod in OCS tapering and FVC[2]. These findings prompted the current post hoc analysis, in which we leveraged the opportunity to determine a concentration of efzofitimod that effectively inhibited granuloma formation *in vitro* in order to identify which dose(s) of the drug used in the clinical trial resulted in serum concentrations that might similarly inhibit granulomatous inflammation *in vivo*, and thereby further evaluate its therapeutic potential in patients with sarcoidosis.

The *in vitro* human granuloma formation assay we employed is an established model that recapitulates morphologic and molecular features of granulomas in patients with sarcoidosis[3,5,6] . The assay has been used previously to determine the concentration of another agent in development for treatment of sarcoidosis to inform the dose range to be tested in a clinical trial[4]. In the current study, we found that efzofitimod at 300 nM (19 µg/mL) significantly inhibited granuloma formation *in vitro* (Figure 1). Pharmacokinetic data from the clinical trial indicated that the Cavg for the 3 mg/kg dose cohort (18 µg/mL) was similar to, and for the 5 mg/kg cohort (24 µg/mL) above, the effective concentration *in vitro*. On the other hand, the Cavg for the 1 mg/kg cohort was well below the effective concentration *in vitro*. This provided the rationale to pool the 3 and 5 mg/kg cohorts as the Therapeutic group, and the 1 mg/kg cohort with the placebo cohort as the Subtherapeutic group, then to compare outcomes in the two groups.

Time-to-first-relapse after an initial successful OCS taper and change in FVC over the course of the study were pre-specified secondary and exploratory endpoints in the phase 1b/2a clinical trial, respectively. Our analysis shows a highly significant reduction in relapses after OCS taper in the Therapeutic group (7.7%) compared to the Subtherapeutic group (54.4%) and a markedly longer time-to-first-relapse in the Therapeutic group (Figure 2, Table 2). Regarding changes in pulmonary function, FVC increased in the Therapeutic group and decreased in the Subtherapeutic group over the course of the trial, such that the mean FVC was significantly (180 mL) greater in the Therapeutic group than the Subtherapeutic group at the end of the trial (Figure 3b, Table 2). FEV_1 similarly increased in the Therapeutic group and decreased in the Subtherapeutic group, although the difference at the end of the trial (86 mL) did not reach statistical significance (Figure 4b, Table 2). Notably, it was the pooling of treatment cohorts into Therapeutic and Subtherapeutic groups based on results from the *in vitro* granuloma assay that allowed us to glean the therapeutic benefit of efzofitimod with respect to these secondary endpoints in the clinical trial.

The European Respiratory Society (ERS) clinical practice guidelines recommend corticosteroids as first-line therapy for treatment of symptomatic pulmonary sarcoidosis to improve or preserve pulmonary function and quality of life[10]. The ERS guidelines also recommend methotrexate or other non-steroid immunomodulators in patients who have continued disease activity despite corticosteroid therapy to improve or preserve pulmonary function[10]. Conversely, discontinuing corticosteroids has been associated with clinical worsening in pulmonary

sarcoidosis[14]. In the phase 1b/2a efzofitimod clinical trial[1], all patients were treated with corticosteroids and 14/37 (38%) were on methotrexate or another non-steroid immunomodulator at study entry (Table 2). In this context, the reduction in relapses and improvement in FVC and FEV_1 in the Therapeutic group at the same time that corticosteroids were tapered strongly suggests a therapeutic benefit of efzofitimod at the 3 and 5 mg/kg doses.

While corticosteroids have been the cornerstone of sarcoidosis therapy for decades, long-term treatment, especially at high doses, is associated with substantial toxicity and decreased quality of life[15-17]. Therefore, the ability of a new therapy to maintain disease control while discontinuing or lowering the dose of corticosteroids is an outcome of clinical importance that is also meaningful to patients. The post hoc analysis presented here suggests that therapeutic doses of efzofitimod can be effective in achieving this goal.

To assess the effect of efzofitimod on lung function, we focused on three pulmonary function parameters: FVC, FEV1 and DLCO. On average, each of these was mildly to moderately decreased in our study population (Table 1). These results are similar to those recently reported from a large tertiary sarcoidosis specialty center[18]. In that study, 56% of patients with pulmonary sarcoidosis had abnormal lung function. Of these, 47% had restrictive impairment, 22% had obstructive impairment, 16% had combined restriction and obstruction, and 15% had an isolated reduction in DLCO[18]. Consistent with the observation that restrictive impairment is the most common physiologic abnormality in pulmonary sarcoidosis, FVC is the most frequently reported pulmonary function parameter in clinical studies, and the one most likely to improve in response to therapy[19]. Likewise, efzofitimod treatment had the largest impact on FVC, which increased in the Therapeutic group and decreased in the Subtherapeutic group, leading to a statistically significant difference of 180 mL by the end of the 24-week study (Figure 3, Table 2). This degree of difference in FVC is large by comparison with the effect of infliximab in the landmark randomized, placebo-controlled clinical trial with that agent[20], and in the same range as the increase in FVC seen in two uncontrolled case series in which infliximab was used at higher doses[21,22]. Although not approved for use in sarcoidosis by either the U.S. Food and Drug Administration or the European Medicines Agency, infliximab is now guideline-recommended therapy for severe sarcoidosis that cannot be controlled with corticosteroids and other immunomodulators[10].

Like FVC, FEV1 increased progressively in the Therapeutic group and decreased in the Subtherapeutic group over the course of the 24-week clinical trial, although the difference between the groups did not reach statistical significance (Figure 4, Table 2). Similarly, DLCO

increased in the Therapeutic group and decreased in the Subtherapeutic group, but the difference was not significant at the end of the trial (Table 2). To determine the importance of the trends toward improvement in FEV_1 and DLCO in response to efzofitimod, larger clinical trials are required.

Limitations

Our study has several limitations. First, it has the inherent limitation of a post hoc analysis of data from a prospective study. Mitigating this, however, the outcomes we analysed were pre-specified endpoints in the phase 1b/2a clinical trial. In addition, we used the same mITT approach and data handling rules specified in advance and applied in the primary report of results from the clinical trial[1].

Second, while equating the concentration of efzofitimod that decreased granuloma formation in the in *vitro* assay to a serum concentration expected to be therapeutically efficacious in sarcoidosis patients is a rational approach, whether activity *in vitro* correlates with the ability to suppress granulomatous inflammation *in vivo* has not been established. Importantly, our finding that relapses decreased and pulmonary function improved while corticosteroids were tapered in the Therapeutic group suggests that an *in vitro-in vivo* correlation may indeed exist.

Third, there are no universally accepted criteria for relapse in pulmonary sarcoidosis, and various studies have defined relapse differently[23-26]. Most authors consider recurrent symptoms, worsening radiographic findings, and/or decline in pulmonary function occurring within 1 to 12 months after medication taper as markers of disease relapse[23-26]. Since radiographic findings may not worsen and pulmonary function may not decline before symptoms increase as corticosteroids are tapered, particularly when other immunomodulatory therapy is in place, our study focused on recurrence of symptoms as the primary indicator of relapse or disease progression. In the double blind clinical trial, investigators at each study site were required to adjudicate whether any report of new symptoms was due to worsening sarcoidosis or attributable to another cause in order to ensure that relapses were properly captured, and that resumption of prednisone or a dose increase for non-sarcoidosis reasons were not counted[26].

Finally, the primary endpoint of the phase 1b/2a trial was safety and tolerability of efzofitimod at a range of doses (1, 3 and 5 mg/kg), while effects on steroid tapering and on pulmonary function were secondary and exploratory endpoints, respectively. The sample size was therefore small, such that the study was not powered to show therapeutic efficacy. For this

reason, the favourable effects of efzofitimod on relapse rates and pulmonary function in the Therapeutic group (pooled 3 and 5 mg/kg dose cohorts) shown in our post hoc analysis must be considered preliminary evidence of clinical benefit. An ongoing phase 3 trial of efzofitimod at 3 and 5 mg/kg versus placebo over 48 weeks has a targeted enrolment of 264 subjects with corticosteroid-requiring pulmonary sarcoidosis[27]. Hopefully, this study will provide definitive evidence as to the efficacy of efzofitimod in preventing relapses, preserving pulmonary function, and other clinical outcomes while tapering corticosteroids.

Conclusion

In conclusion, using an established assay with cultured human PBMCs, we identified a concentration of efzofitimod at or above which granuloma formation was inhibited *in vitro*. This, in combination with pharmacokinetic data from the phase 1b/2a study, allowed us to designate patients treated with efzofitimod at 3 and 5 mg/kg as having received therapeutic doses (the Therapeutic group) and those dosed at 1 mg/kg as having received a subtherapeutic dose (combined with the placebo cohort as the Subtherapeutic group). Our post hoc analysis revealed that over half of patients in the Subtherapeutic group relapsed after a successful corticosteroid taper, while fewer than 10% of those in the Therapeutic group relapsed. The analysis also showed that efzofitimod at therapeutic doses favourably impacted pulmonary function: FVC increased significantly, and there were trends toward improvement in FEV1 and DLCO in the Therapeutic group compared to the Subtherapeutic group. These findings build upon the results reported from the primary analysis of the phase 1b/2a trial[1] and subsequent exposure-response analysis[2] supporting the efficacy of efzofitimod in allowing corticosteroid tapering and improving quality of life measures and stability of pulmonary function in patients with pulmonary sarcoidosis. Ultimately, however, the true therapeutic benefit of efzofitimod as a novel biologic therapy for sarcoidosis will depend on larger randomized controlled studies, such as the phase 3 clinical trial currently underway.

Conflict of Interest

Ogugua N. Obi has received support for the conduct of clinical trials from aTyr Pharma, Novartis, Kinevant, and Xentria, has served as consultant for CSL-Behring, and Xentria, served on the scientific advisory board (SAB) of the Foundation of Sarcoidosis Research, and currently serves on the SAB of the Ann Theodore Foundation. Elliott D. Crouser has received grant support from aTyr Pharma, Xentria, 23&ME, Star Therapeutics, Milken Institute/Ann Theodore Foundation, and the Foundation for Sarcoidosis Research, and has served as a consultant to Boehringer Ingelheim, SarcoMedUSA, and Merck. Mark W. Julian and Landon W. Locke have no conflict of interest to declare. Abhijeeth Chandrasekaran, Pavithra Ramesh, Nelson Kinnersley, and Vis Niranjan declare that support provided by them was contracted and funded by aTyr Pharma. Robert P. Baughman has received support for clinical trials from aTyr Pharma, Celgene, Actelion, Genentech, Gilead, Bellerophon, Bayer, Mallinckrodt, the Foundation for Sarcoidosis Research and served as consultant to Astra Zeneca, Actelion, aTyr, Kinevant, Xentria, Mallinckrodt, Boehringer Ingelheim, Foresee Pharmaceuticals, and the Ann Theodore Foundation, and been on speaker's bureau for Mallinckrodt. Daniel A. Culver has received grant support and/or consulting fees from the Ann Theodore Foundation, aTyr Pharma, Kinevant, Molecure, Mallinckrodt, Merck, Boehringer-Ingelheim, Foree Pharmaceuticals, and the Foundation for Sarcoidosis Research. Peter H. S. Sporn has received support for the conduct of clinical trials from aTyr Pharma, Novartis, Xentria and the Foundation for Sarcoidosis Research, and has served as a consultant to ANI Pharmaceuticals.

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Table 1. Patient demographics, baseline disease characteristics and baseline immunosuppressive therapy.

	Subtherapeutic (N=20)	**Therapeutic (N=17)**
Patient Demographics		
Age, years (mean; SD), ≥ 65	53.3 (10.4), 1	51.2 (10.0), 2
Sex (Female), N (%)	11 (55)	9 (53)
Race (White/African American), N	14/6	9/8
Baseline Disease Characteristics, Mean (SD)		
FVCpp, %	73.7 (11.5)	83.8 (12.7)
FVC, mL	2816 (739)	3396 (1018)
FEV1pp, %	65.2 (17.0)	77.5 (15.6)
FEV1, mL	1942.3 (546.8)	2502.6 (915.9)
DLCOpp, %	62 (20)	67 (20)
Duration of Disease, years	5.5 (4.7)	6.9 (7.9)
Baseline Dyspnea Index Score	4.6 (1.8)	6.9 (2.7)
Baseline Therapy, N (%)		
Prednisone-equivalent dose, mg/day		
20-25	4 (20)	4 (24)
15 to <20	2 (10)	5 (29)
10 to <15	14 (70)	8 (47)
Mean	12.5	14.1
Non-steroid Immunomodulator	9 (45)	5 (29)
Methotrexate	6	3
Azathioprine	2	1
Hydroxychloroquine	1	0
Leflunomide	0	1

Table 2. Time-to-first-relapse and pulmonary function by treatment group.

	Subtherapeutic	Therapeutic	Treatment Effect	P value
	N = 20	**N = 17**		
Relapses				
Subjects tapered, N	17	15		
Subjects with relapse, N (%)	8 (54.4)	1 (7.7)		
Median time-to-first-relapse, days	126	NE	NE	0.017
FVC				
N	**13**	**13**		
Week 24 mean, mL (SD)	2537 (818)	3615 (1253)		
Week 24 adjusted mean[1]	2985	3165	180	0.035
FEV_1				
N	**13**	**13**		
Week 24 mean, mL (SD)	1734 (961)	2685 (1047)		
Week 24 adjusted mean[1]	2146	2232	86	0.196
DLCOpp				
Baseline				
N	**19**	**14**		
Percent, mean (SD)	62 (20)	67 (20)		
Week 12				
N	**17**	**9**		
Percent, mean (SD)	61 (20)	68 (26)		
Percent, adjusted mean[2]	57.9	67.2	9.3	0.013
Week 20				
N	**12**	**11**		
Percent, mean (SD)	60 (18)	68 (23)		
Percent, adjusted mean[2]	61.2	63.5	2.3	0.630
Week 24				
N	**10**	**11**		
Percent, mean (SD)	53 (15)	70 (23)		
Percent, adjusted mean[2]	57.7	65.1	7.4	0.104

NE = not estimable

[1] Based on a slopes analysis adjusted for covariates.

[2] Based on MMRM adjusted for covariates.

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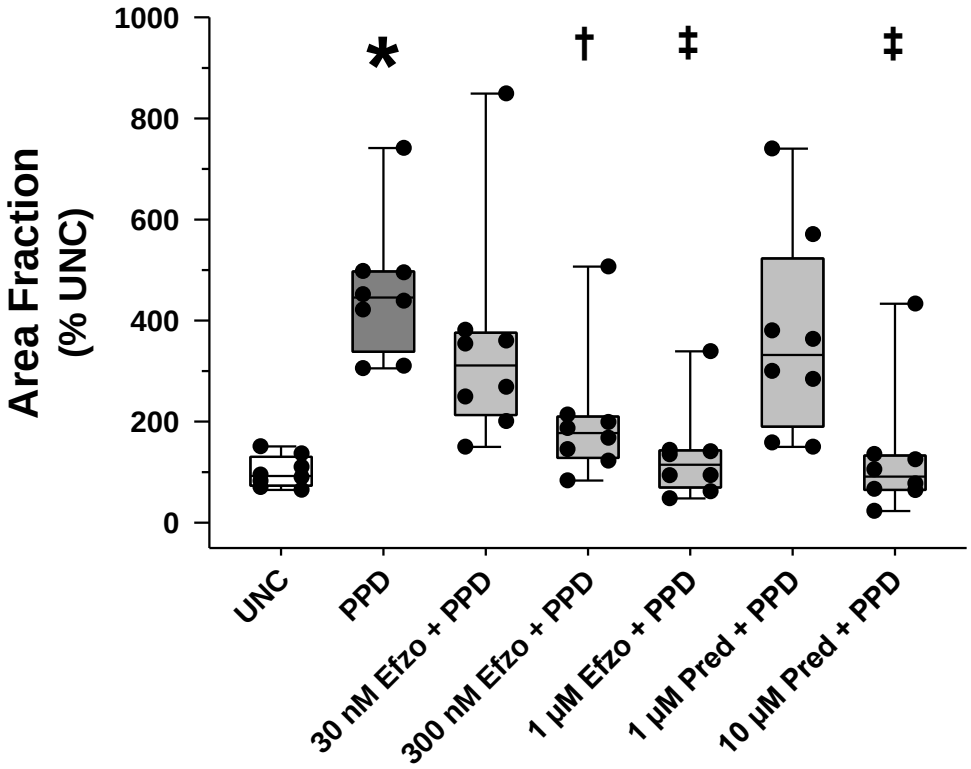
Area Fraction
(% UNC)
1000
800
600
400
200
0
*
†
‡
‡
UNC
PPD
30 nM Efzo + PPD
300 nM Efzo + PPD
1 µM Efzo + PPD
1 µM Pred + PPD
10 µM Pred + PPD

Figure 1

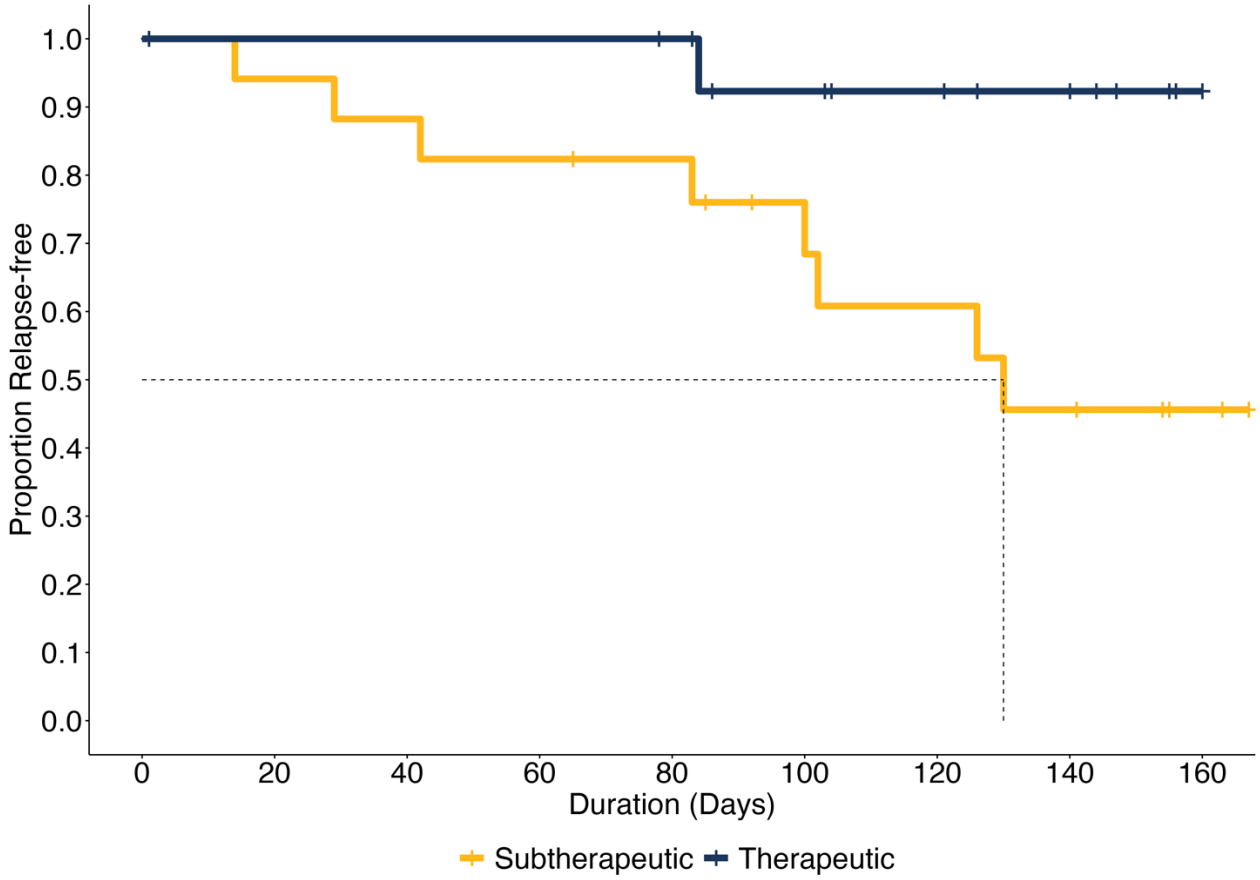
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
Proportion Relapse-free
0
20
40
60
80
100
120
140
160
Duration (Days)
Subtherapeutic
Therapeutic

Figure 2

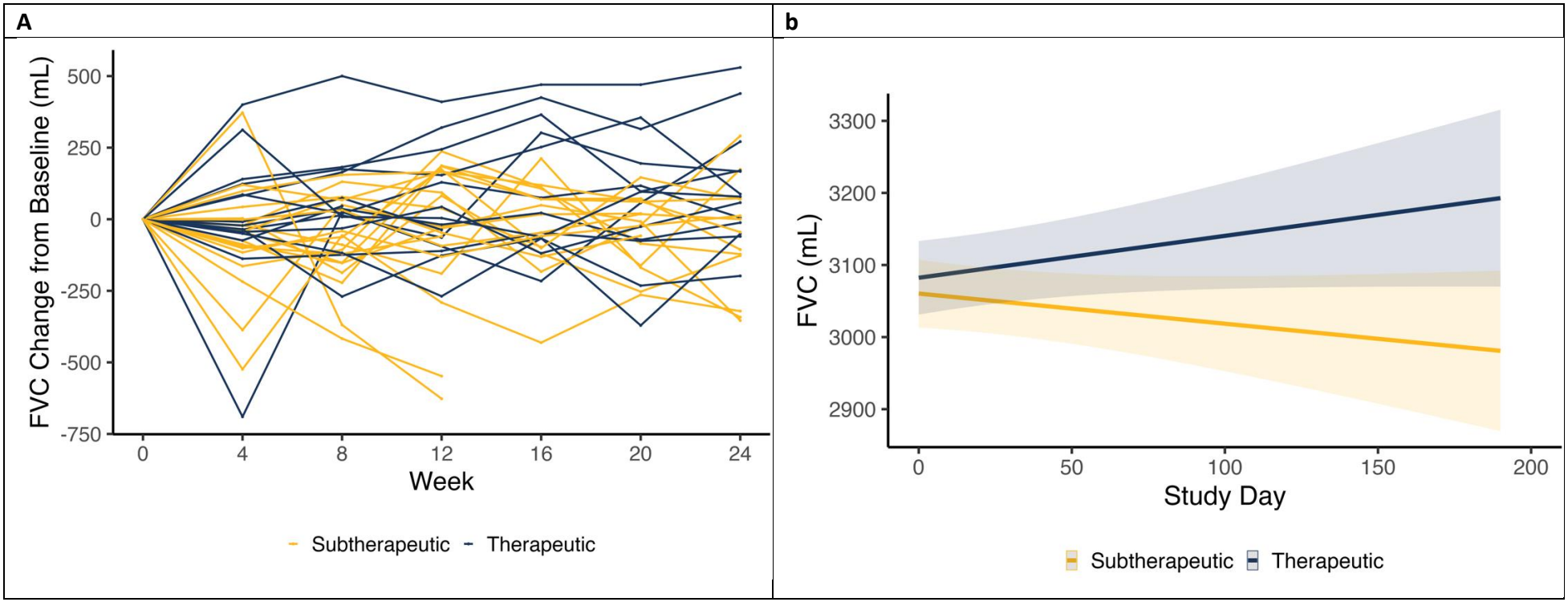
A
FVC Change from Baseline (mL)
500
250
0
-250
-500
-750
0
4
8
12
16
20
24
Week
Subtherapeutic
Therapeutic
b
FVC (mL)
3300
3200
3100
3000
2900
0
50
100
150
200
Study Day
Subtherapeutic
Therapeutic

Figure 3

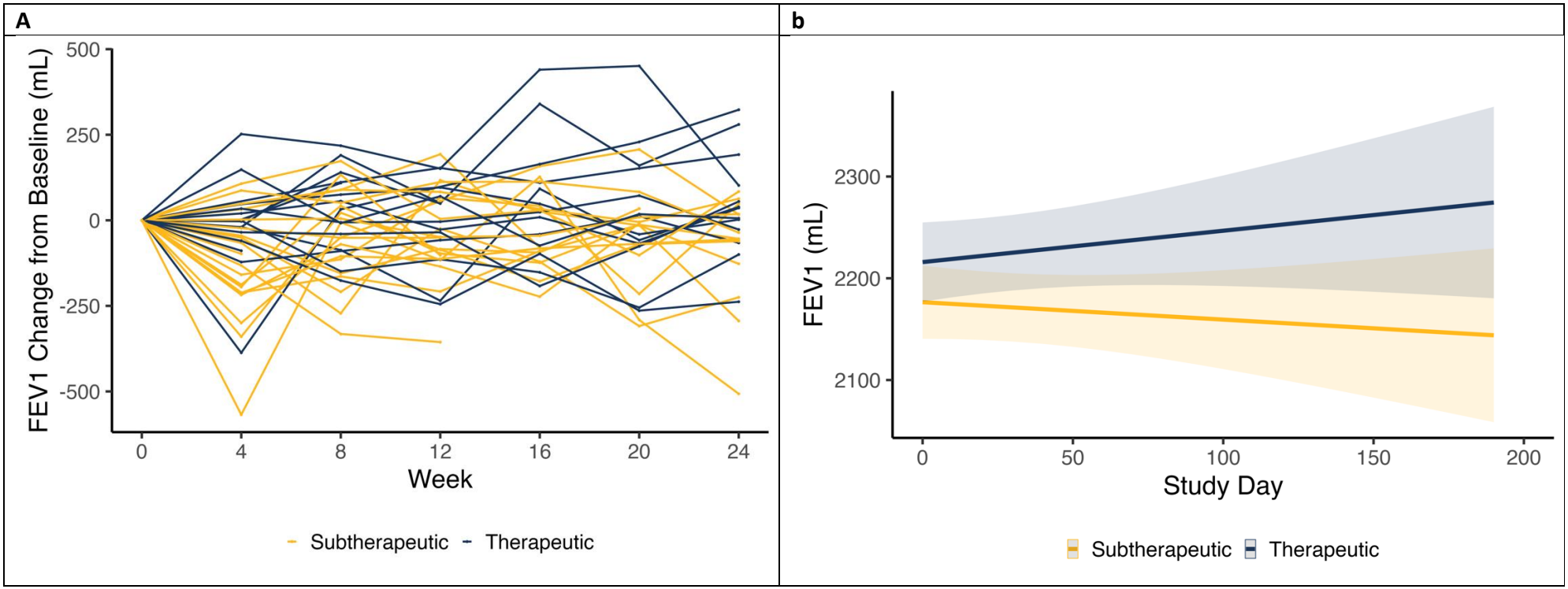
A
FEV1 Change from Baseline (mL)
500
250
0
-250
-500
0
4
8
12
16
20
24
Week
Subtherapeutic
Therapeutic
b
FEV1 (mL)
2300
2200
2100
0
50
100
150
200
Study Day
Subtherapeutic
Therapeutic

Figure 4

Supplemental Table 1: Sarcoid patient donor demographics for the *in vitro* study.

Group	Age (years)	Sex (M/F)	Race (W/B/O)*	Ethnicity (H/NH†)	Scadding CXR Stage	Extra-pulmonary
Sarcoidosis (n = 8)	45	F	B	NH	III	Yes
	63	M	B	NH	IV	Yes
	52	M	W	NH	II	No
	66	F	B	NH	II	Yes
	59	M	W	NH	II	Yes
	59	F	B	NH	IV	Yes
	64	F	W	NH	I	No
	58	M	W	NH	III	Yes
Mean ± SD or Ratio	**58.3 ± 6.9**	**4/4**	**4/4/0**	**0/8**		

*White/Black/Other
†Hispanic/Non-Hispanic